

Office Action Application No. 09/943,002  
Amendment dated May 12, 2005  
Reply to Office Action of November 12, 2004

**Amendments to the Claims:**

This listing of claims will replace all prior versions, and listings, of claims in the application.

**Listing of Claims:**

Claim 1 (canceled)

Claim 2 (canceled)

Claim 3 (canceled)

Claim 4 (currently amended) A protein characterized as:

having a molecular weight of about 15,000,

having less than ~~about~~ 150 amino acid residues,

having one transmembrane domain,

having one ~~relatively small~~ intracellular domain,

having an a-relatively small extracellular domain, wherein said extracellular

domain contains an alpha helix motif, and

being substantially ~~relatively~~ non-immunogenic in a mammal,

wherein said protein is further characterized as lacking:

signal peptide, and

N-linked glycosylation signals.

Claim 5 (currently amended) The protein of claim 4 having an amino acid sequence substantially the same as set forth in SEQ ID NO: 14 ~~[BRV]~~.

Claim 6 (currently amended) The protein of claim 4 having the amino acid sequence set forth in SEQ ID NO: 14 ~~[BRV]~~.

Claim 7 (withdrawn) An antibody raised against the protein of claim 43.

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Claim 8 (withdrawn) An antibody raised against the protein of claim 4.

Claim 9 (withdrawn) An isolated nucleic acid encoding the protein of claim 43.

Claim 10 (withdrawn) An isolated nucleic acid according to claim 9 having a contiguous nucleotide sequence substantially the same as:

nucleotides 25-1607 of SEQ ID NO: 1 [ARV1],

nucleotides 25-1607 of SEQ ID NO: 5 [ARV2],

nucleotides 27-1579 of SEQ ID NO: 9 [NBV], or

variations thereof which encode the same amino acid sequence, but employ different codons for some of the amino acids, or splice variant nucleotide sequences thereof.

Claim 11 (withdrawn) An isolated and purified nucleic acid, or functional fragment thereof encoding the protein of claim 43, wherein the nucleic acid is selected from the group consisting of:

(a) DNA encoding the amino acid sequence set forth in SEQ ID NO: 2, SEQ ID NO: 6 or SEQ ID NO: 10, or

(b) DNA that hybridizes to the DNA of (a) under moderately stringent conditions, wherein said DNA encodes a biologically active fusion protein, or

(c) DNA degenerate with respect to either (a) or (b) above, wherein said DNA encodes a biologically active fusion protein.

Claim 12 (withdrawn) An isolated nucleic acid according to claim 9 operatively associated with an inducible promoter.

Claim 13 (withdrawn) An isolated nucleic acid encoding the protein of claim 4.

Claim 14 (withdrawn) The isolated nucleic acid of claim 13 having a contiguous nucleotide sequence substantially the same as:

nucleotides 25-832 of SEQ ID NO: 13 [BRV], or

variations thereof which encode the same amino acid sequence, but employ different codons for some of the amino acids, or splice variant nucleotide sequences thereof.

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**Claim 15 (withdrawn)** An isolated and purified nucleic acid, or functional fragment thereof encoding the protein of claim 4, wherein the nucleic acid is selected from the group consisting of:

- (a) DNA encoding the amino acid sequence set forth in SEQ ID NO: 14, or
- (b) DNA that hybridizes to the DNA of (a) under moderately stringent conditions, wherein said DNA encodes a biologically active fusion protein, or
- (c) DNA degenerate with respect to either (a) or (b) above, wherein said DNA encodes a biologically active fusion protein.

**Claim 16 (withdrawn)** The isolated nucleic acid of claim 13 operatively associated with an inducible promoter.

**Claim 17 (withdrawn)** A cell containing the protein of claim 43.

**Claim 18 (withdrawn)** The cell containing the protein of claim 4.

**Claim 19 (withdrawn)** The cell containing the nucleic acid of claim 9.

**Claim 20 (withdrawn)** The cell containing the nucleic acid of claim 12.

**Claim 21 (withdrawn)** The cell containing the nucleic acid of claim 13.

**Claim 22 (withdrawn)** The cell containing the nucleic acid of claim 16.

**Claim 23 (withdrawn)** Liposomes containing the protein of claim 43.

**Claim 24 (withdrawn)** Liposomes containing the protein of claim 4.

**Claim 25 (withdrawn)** Liposomes containing the nucleic acid of claim 9.

**Claim 26 (withdrawn)** Liposomes containing the nucleic acid of claim 13.

**Claim 27 (withdrawn)** A method for producing the protein of claim 43, said method comprising the step of expressing a nucleic acid encoding said protein in a suitable host.

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**Claim 28 (withdrawn)** A method for producing the protein of claim 4, said method comprising the step of expressing a nucleic acid encoding said protein in a suitable host.

**Claim 29 (canceled)**

**Claim 30 (canceled)**

**Claim 31 (canceled)**

**Claim 32 (canceled)**

**Claim 33 (canceled)**

**Claim 34 (canceled)**

**Claim 35 (canceled)**

**Claim 36 (canceled)**

**Claim 37 (canceled)**

**Claim 38 (canceled)**

**Claim 39 (canceled)**

**Claim 40 (canceled)**

**Claim 41 (withdrawn)** An isolated nucleic acid fragment useful as a hybridization probe, wherein said fragment comprises at least 14 contiguous nucleotides of the nucleic acid according to Claim 9, and wherein said fragment is labeled with a detectable substituent.

**Claim 42 (withdrawn)** An isolated nucleic acid fragment useful as a hybridization probe, wherein said fragment comprises at least 14 contiguous nucleotides of the nucleic acid according to Claim 13, and wherein said fragment is labeled with a detectable substituent.

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Claim 43 (withdrawn) An isolated protein which:

- (a) is a membrane fusion protein;
- (b) comprises a transmembrane domain; and
- (c) has at least 33% amino acid sequence identity to a protein which:
  - (i) is encoded by a polynucleotide from the genome of Reoviridae;
  - (ii) is a membrane fusion protein, and
  - (iii) has a molecular weight of about 11 kDa.

Claim 44 (withdrawn) The isolated protein of claim 43 which:

- (a) is encoded by a polynucleotide from the genome of Reoviridae; and
- (b) has molecular weight of about 11 kDa.

Claim 45 (withdrawn) The protein of claim 43 which has less than 100 amino acids or has about 100 amino acids.

Claim 46 (withdrawn) The protein of claim 43 which contains a cluster of positive amino acid residues, wherein the cluster is located on the C-terminal side of the transmembrane domain and comprises at least 4 positive residues within the 20 residues flanking the transmembrane domain at the C-terminal side.

Claim 47 (withdrawn) The protein of claim 43 which lacks a signal peptide.

Claim 48 (withdrawn) The protein of claim 43 which contains 4 cysteine residues at conserved positions relative to SEQ ID NO:2 (ARV1); SEQ ID NO:6 (ARV2); and SEQ ID NO:10 (NBV).

Claim 49 (withdrawn) The isolated protein of claim 43 which has at least 33% amino acid sequence identity to a polypeptide selected from the group consisting of:

- (a) a polypeptide of SEQ ID NO:2 (ARV1);
- (b) a polypeptide of SEQ ID NO:6 (ARV2); and
- (c) a polypeptide of SEQ ID NO:10 (NBV).

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Claim 50 (withdrawn) The protein of claim 49 which has at least 33% amino acid sequence identity to the polypeptide of SEQ ID NO:2 (ARV1).

Claim 51 (withdrawn) The protein of claim 49 which has at least 33% amino acid sequence identity to the polypeptide of SEQ ID NO:6 (ARV2).

Claim 52 (withdrawn) The protein of claim 49 which has at least 33% amino acid sequence identity to the polypeptide of SEQ ID NO:10 (NBV).

Claim 53 (withdrawn) The protein of claim 49 which comprises a cluster of positive amino acid residues, wherein the cluster is located on the C-terminal side of the transmembrane domain and comprises at least 4 positive residues within the 20 residues flanking the transmembrane domain at the C-terminal side.

Claim 54 (withdrawn) The protein of claim 49 which lacks a signal peptide.

Claim 55 (withdrawn) The protein of claim 49 which comprises 4 cysteine residues at conserved positions relative to SEQ ID NO:2 (ARV1); SEQ ID NO:6 (ARV2); and SEQ ID NO:10 (NBV).

Claim 56 (withdrawn) An isolated protein comprising the sequence selected from the group consisting of: SEQ ID NO:2(ARV1), SEQ ID NO:6(ARV2) and SEQ ID NO:10(NBV).

Claim 57 (currently amended) An isolated protein which:

- (a) induces ~~[[is a]]~~ membrane fusion protein;
- (b) comprises a transmembrane domain;
- (c) has a molecular weight of about 15 kDa; and
- (d) is encoded by a polynucleotide from the genome of Reoviridae.

Claim 58 (withdrawn) A method to promote membrane fusion, said method comprising the step of contacting the membranes to be fused with the protein of claim 43 for a time and under conditions effective to promote membrane fusion.

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Claim 59 (withdrawn) A method to promote membrane fusion, said method comprising the step of contacting the membranes to be fused with the protein of claim 57 for a time and under conditions effective to promote membrane fusion.

Claim 60 (withdrawn) A method to promote membrane fusion, said method comprising the step of contacting the membranes to be fused with a membrane fusion protein for a time and under conditions effective to promote membrane fusion, wherein the membrane fusion protein is encoded by a polynucleotide of the genome of a fusogenic member of the family Reoviridae or is substantially the same as the membrane fusion protein encoded by a polynucleotide of the genome of a fusogenic member of the family Reoviridae.

Claim 61 (withdrawn) The method of claim 60 wherein the fusogenic member of the family Reoviridae is selected from: ARV, NBV and BRV.

Claim 62 (withdrawn) The method of claim 60 wherein the membranes are cell membranes, liposome membranes or proteoliposome membranes.

Claim 63 (withdrawn) The method of claim 60 wherein the membranes are the cell membrane of an immortalized myeloma cell and the cell membrane of a primary B cell or T cell.

Claim 64 (withdrawn) The method of claim 63 wherein the immortalized myeloma cell is human or mouse, and wherein the primary B cell or T cell is a purified spleen cell from an immunized mammal.

Claim 65 (withdrawn) The method of claim 60 wherein the membranes to be fused are the cell membrane of an immortalized myeloma cell and the cell membrane of a primary B cell or T cell, and wherein the membrane fusion protein has an amino acid sequence selected from any one of: SEQ ID NOs: 2, 6, 10, 14, and substantially the same sequences thereof.

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Claim 66 (withdrawn) A method to promote membrane fusion between a first membrane and a second membrane, said method comprising:

- (a) introducing a fusogenic protein, in an amount sufficient to effect membrane fusion, into the first membrane; and then,
- (b) contacting the second membrane with the first membrane for a time and under conditions effective to promote membrane fusion between the first membrane and the second membrane;

wherein the first membrane is selected from the group consisting of:

- (i) a liposome membrane;
- (ii) a proteoliposome membrane; and
- (iii) a membrane of a cell;

and wherein the fusogenic protein either:

- (i) is encoded by a polynucleotide of the genome of Reoviridae; and
- (ii) has a molecular weight of about 11 kDa; and
- (iii) is less than 100 amino acids or is about 100 amino acids;

or wherein the fusogenic protein:

- (i) is encoded by a polynucleotide of the genome of Reoviridae; and
- (ii) has a molecular weight of about 15 kDa; and
- (iii) is less than 150 amino acids or is about 150 amino acids.

Claim 67 (withdrawn) The method according to claim 66 wherein the first membrane is the liposome membrane or the proteoliposome membrane.

Claim 68 (withdrawn) The method according to claim 67, wherein the step of introducing the fusogenic protein comprises incorporating the fusogenic protein into the liposome membrane or the proteoliposome membrane.



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Claim 69 (withdrawn) The method according to claim 66 wherein the first membrane is the cell membrane.

Claim 70 (withdrawn) The method according to claim 69, wherein the step of introducing the fusogenic protein comprises the step of introducing into the cell an expression vector comprising a polynucleotide which encodes the fusogenic protein, wherein the vector is free of full-length reovirus genome.

Claim 71 (withdrawn) A method to promote membrane fusion between a first membrane and a second membrane, said method comprising contacting the first or second membrane or both membranes with an effective amount of a protein-liposome complex for a time and under conditions effective to promote membrane fusion between the first membrane and the second membrane, wherein the protein-liposome complex contains a fusogenic protein; and

wherein the fusogenic protein either:

- (i) is encoded by a polynucleotide of the genome of Reoviridae;
- (ii) has a molecular weight of about 11 kDa; and
- (iii) is less than 100 amino acids or is about 100 amino acids;

or wherein the fusogenic protein:

- (i) is encoded by a polynucleotide of the genome of Reoviridae;
- (ii) has a molecular weight of about 15 kDa; and
- (iii) is less than 150 amino acids or is about 150 amino acids.

Claim 72 (withdrawn) The method according to claim 66 wherein the fusogenic protein comprises an amino acid sequence selected from any one of: SEQ ID NOs: 2, 6, 10 and 14.

Claim 73 (withdrawn) The method according to claim 67 wherein the fusogenic protein comprises an amino acid sequence selected from any one of: SEQ ID NOs: 2, 6, 10 and 14.

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Claim 74 (withdrawn) The method according to claim 68 wherein the fusogenic protein comprises an amino acid sequence selected from any one of: SEQ ID NOs: 2, 6, 10 and 14.

Claim 75 (withdrawn) The method according to claim 69 wherein the fusogenic protein comprises an amino acid sequence selected from any one of: SEQ ID NOs: 2, 6, 10 and 14.

Claim 76 (withdrawn) The method according to claim 70 wherein the fusogenic protein comprises an amino acid sequence selected from any one of: SEQ ID NOs: 2, 6, 10 and 14.

Claim 77 (withdrawn) The method according to claim 71 wherein the fusogenic protein comprises an amino acid sequence selected from any one of: SEQ ID NOs: 2, 6, 10 and 14.